

seq list

SEQUENCE LISTING

<110> Ago, Hideo
Miyano, Masashi
Adachi, Tsuyoshi

<120> HCV Polymerase Suitable for Crystal
Structure Analysis and Method for Using the Enzyme

<130> SHIM007

<140> 09/608,713

<141> 2000-06-30

<150> 11-188630

<151> 1999-07-02

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<151> 1999-07-07

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<212> FRT

<213> Hepatitis C Virus

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<221> CDS

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<223> DNA encoding fusion protein consistin of a portion
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<221> misc_feature

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<223> n = A,T,C or G

<221> misc_feature

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<223> n = A,T,C or G

<223> DNA encoding fusion protein consisting of a
portion of HCV polymerase and histidine tag at the
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<223> n = A,T,C or G

<221> misc_feature

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<223> n = A,T,C or G

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ctg	ccc	atc	aac	ggg	ttg	agc	aac	tct	ttg	ctg	aag	ugu	sry	sur	asn	144
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<223> Xaa = Any Amino Acid

<223> DNA encoding fusion protein consisting of a
portion of HCV polymerase and histidine tag at the
C-terminus

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		130				135					140				
Asp	Ala	Pro	Thr	Xaa	Xaa	Xaa	Cys	Asn	Xaa	Xaa	Xaa	Xaa	Ser	Ala	Lys
145				150						155				160	
Ser	Lys	Phe	Gly	Tyr	Gly	Ala	Lys	Asp	Val	Arg	Asn	Leu	Xaa	Xaa	Xaa

seq list																		
				165					170					175				
Xaa	Xaa	Xaa	Asn	Xaa	Xaa	Xaa	Xaa	Ser	Ser	Lys	Ala	Val	Asn	His	Ile			
			180					185					190					
His	Ser	Val	Trp	Lys	Asp	Leu	Leu	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		
		195					200						205					
Xaa	Leu	Lys	Thr	Leu	His	Gln	Leu	Thr	Pro	Pro	Ser	Trp	Gln	Lys	Met			
	210					215					220							
Arg	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Phe	Leu	Cys	Pro	Thr			
225					230					235					240			
Arg	Glu	Arg	Arg	Pro	Ala	Ser	Pro	Pro	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		
				245					250					255				
Xaa	Xaa	Lys	Val	Ser	Tyr	Ser	Gln	Ile	Trp	Glu	Ser	Val	Tyr	Ala	Arg			
		260					265						270					
Arg	Trp	Pro	Xaa	Xaa	Xaa	Xaa	Lys	Xaa	Xaa	Xaa	Xaa	Asn	Tyr	Asp	Val			
	275						280					285						
Val	Ser	Thr	Leu	Pro	Gln	Val	Val	Met	Gly	Ser	Ser	Tyr	Xaa	Xaa	Xaa			
	290				295						300							
Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Gly	Phe	Gln	Tyr	Ser	Pro	Gly	Gln			
305					310					315					320			
Arg	Val	Glu	Phe	Leu	Val	Asn	Thr	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		
				325				330						335				
Xaa	Xaa	Xaa	Gly	Asn	Gln	Arg	Lys	Thr	Pro	Trp	Ala	Phe	His	Met	Thr			
			340					345					350					
Leu	Ala	Val	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Arg		
		355					360						365					
Leu	Asn	Gly	His	Arg	Glu	Arg	His	Pro	Cys	Gly	Val	Asn	Xaa	Xaa	Xaa			
	370					375					380							
Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Cys	Xaa	Thr	Asn	Val	Val	Thr	Trp	Pro	Pro			
385					390					395					400			
Lys	Pro	Asp	Arg	Pro	Asn	Arg	Xaa	Xaa	Xaa	Xaa	Xaa	Lys	Xaa	Xaa	Xaa			
				405				410						415				
Xaa	His	Arg	Ala	Ala	Leu	Tyr	Arg	Gly	Ser	Ser	Asp	Phe	Lys	Arg	Arg			
			420					425					430					
Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Arg	Thr	Ala	Val	Ile	Ala			
		435					440					445						
Gly	Ala	Ala	Arg	Ala	Ala	Cys	Arg	Leu	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa			
	450				455						460							
Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Ser	Cys	Gly	Asn	Thr	Leu	Thr	Cys	Tyr	Leu			
465					470					475					480			
Lys	Ala	Ser	Ala	Ala	Cys	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa			
				485				490						495				
Lys	Xaa	Xaa	Ser	Cys	Glu	Ala	Pro	Gly	Leu	His	Asp	Ala	Arg	Glu	Arg			
			500					505					510					
Arg	Arg	Gln	Glu	Asn	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Ser	Ser			
		515					520						525					
Leu	Ser	Val	Lys	Ala	Arg	Glu	Pro	Lys	Arg	Thr	Arg	Arg	Ala	Xaa	Xaa			
	530					535					540							
Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Lys	Xaa	Leu	Arg	Val	Phe	Thr	Glu	Ala	Met			

seq list

545 550 555 560
 Thr Arg Tyr Ser Ala Pro Pro Gly Xaa Xaa
 565 570

<210> 4
 <211> 579
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> DNA encoding fusion protein consisting of a
 portion of HCV polymerase and histidine tag at the
 C-terminus

<400> 4
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 1 5 10 15
 Ala Glu Glu Ser Lys Leu Pro Ile Asn Ala Leu Ser Asn Ser Leu Leu
 20 25 30
 Arg His His Asn Met Val Tyr Ala Thr Thr Ser Arg Ser Ala Gly Leu
 35 40 45
 Arg Gln Lys Lys Val Thr Phe Asp Arg Leu Gln Val Leu Asp Asp His
 50 55 60
 Tyr Arg Asp Val Leu Lys Glu Met Lys Ala Lys Ala Ser Thr Val Lys
 65 70 75 80
 Ala Lys Leu Leu Ser Val Glu Glu Ala Cys Lys Leu Thr Pro Pro His
 85 90 95
 Ser Ala Lys Ser Lys Phe Gly Tyr Gly Ala Lys Asp Val Arg Asn Leu
 100 105 110
 Ser Ser Lys Ala Val Asn His Ile His Ser Val Trp Lys Asp Leu Leu
 115 120 125
 Glu Asp Thr Val Thr Pro Ile Asp Thr Thr Ile Met Ala Lys Asn Glu
 130 135 140
 Val Phe Cys Val Gln Pro Glu Lys Gly Gly Arg Lys Pro Ala Arg Leu
 145 150 155 160
 Ile Val Phe Pro Asp Leu Gly Val Arg Val Cys Glu Lys Met Ala Leu
 165 170 175
 Tyr Asp Val Val Ser Thr Leu Pro Gln Val Val Met Gly Ser Ser Tyr
 180 185 190
 Gly Phe Gln Tyr Ser Pro Gly Gln Arg Val Glu Phe Leu Val Asn Thr
 195 200 205
 Trp Lys Ser Lys Lys Asn Pro Met Gly Phe Ser Tyr Asp Thr Arg Cys
 210 215 220
 Phe Asp Ser Thr Val Thr Glu Asn Asp Ile Arg Val Glu Glu Ser Ile
 225 230 235 240
 Tyr Gln Cys Cys Asp Leu Ala Pro Glu Ala Arg Gln Ala Ile Lys Ser
 245 250 255

seq list

Leu	Thr	Glu	Arg	Leu	Tyr	Ile	Gly	Gly	Pro	Leu	Thr	Asn	Ser	Lys	Gly	
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Gln	Asn	Cys	Gly	Tyr	Arg	Arg	Cys	Arg	Ala	Ser	Gly	Val	Leu	Thr	Thr	
		275					280					285				
Ser	Cys	Gly	Asn	Thr	Leu	Thr	Cys	Tyr	Leu	Lys	Ala	Ser	Ala	Ala	Cys	
	290					295				300						
Arg	Ala	Ala	Lys	Leu	Gln	Asp	Cys	Thr	Met	Leu	Val	Asn	Gly	Asp	Asp	
305					310					315					320	
Leu	Val	Val	Ile	Cys	Glu	Ser	Ala	Gly	Thr	Gln	Glu	Asp	Ala	Ala	Ser	
			325					330						335		
Leu	Arg	Val	Phe	Thr	Glu	Ala	Met	Thr	Arg	Tyr	Ser	Ala	Pro	Pro	Gly	
			340					345					350			
Asp	Pro	Pro	Gln	Pro	Glu	Tyr	Asp	Leu	Glu	Leu	Ile	Thr	Ser	Cys	Ser	
		355					360					365				
Ser	Asn	Val	Ser	Val	Ala	His	Asp	Ala	Ser	Gly	Lys	Arg	Val	Tyr	Tyr	
	370					375					380					
Leu	Thr	Arg	Asp	Pro	Thr	Thr	Pro	Leu	Ala	Arg	Ala	Ala	Trp	Glu	Thr	
385					390					395					400	
Ala	Arg	His	Thr	Pro	Val	Asn	Ser	Trp	Leu	Gly	Asn	Ile	Ile	Met	Tyr	
			405					410					415			
Ala	Pro	Thr	Leu	Trp	Ala	Arg	Met	Ile	Leu	Met	Thr	His	Phe	Phe	Ser	
			420					425					430			
Ile	Leu	Leu	Ala	Gln	Glu	Gln	Leu	Glu	Lys	Ala	Leu	Asp	Cys	Gln	Ile	
		435					440					445				
Tyr	Gly	Ala	Cys	Tyr	Ser	Ile	Glu	Pro	Leu	Asp	Leu	Pro	Gln	Ile	Ile	
	450					455					460					
Glu	Arg	Leu	His	Gly	Leu	Ser	Ala	Phe	Ser	Leu	His	Ser	Tyr	Ser	Pro	
465					470					475					480	
Gly	Glu	Ile	Asn	Arg	Val	Ala	Ser	Cys	Leu	Arg	Lys	Leu	Gly	Val	Pro	
			485					490						495		
Pro	Leu	Arg	Val	Trp	Arg	His	Arg	Ala	Arg	Ser	Val	Arg	Ala	Arg	Leu	
			500					505					510			
Leu	Ser	Gln	Gly	Gly	Arg	Ala	Ala	Thr	Cys	Gly	Lys	Tyr	Leu	Phe	Asn	
		515					520					525				
Trp	Ala	Val	Lys	Thr	Lys	Leu	Lys	Leu	Thr	Pro	Ile	Pro	Ala	Ala	Ser	
	530					535					540					
Gln	Leu	Asp	Leu	Ser	Gly	Trp	Phe	Val	Ala	Gly	Tyr	Ser	Gly	Gly	Asp	
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Ile	Tyr	His	Ser	Leu	Ser	Arg	Ala	Arg	Pro	Arg	Gly	Ser	His	His	His	
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<211> 30

<212> DNA

<213> Artificial Sequence

seq list

<220>
 <221> primer_bind
 <222> (1)...(30)
 <223> Artificially synthesized primer sequence, 5BNde1FW

<223> DNA encoding fusion protein consisting of a
 portion of HCV polymerase and histidine tag at the
 C-terminus

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30

<210> 6
 <211> 57
 <212> DNA
 <213> Artificial Sequence

<220>
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 <222> (1)...(57)
 <223> Artificially synthesized primer sequence, 5B570HRV

<223> DNA encoding fusion protein consisting of a
 portion of HCV polymerase and histidine tag at the
 C-terminus

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57

<210> 7
 <211> 57
 <212> DNA
 <213> Artificial Sequence

<220>
 <221> primer_bind
 <222> (1)...(57)
 <223> Artificially synthesized primer sequence, 5B552HRV

<223> DNA encoding fusion protein consisting of a
 portion of HCV polymerase and histidine tag at the
 C-terminus

<400> 7
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57

<210> 8

seq list

<211> 57
<212> DNA
<213> Artificial Sequence

<220>
<221> primer_bind
<222> (1)...(57)
<223> Artificially synthesized primer sequence, 5B544HRV

<223> DNA encoding fusion protein consisting of a
portion of HCV polymerase and histidine tag at the
C-terminus

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<210> 9
<211> 67
<212> DNA
<213> Artificial Sequence

<220>
<221> primer_bind
<222> (1)...(67)
<223> Artificially synthesized primer sequence,
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<223> Artificially synthesized primer sequence 5B531HRV

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gaagagg 67

<210> 10
<211> 60
<212> DNA
<213> Artificial Sequence

<220>
<221> primer_bind
<222> (1)...(60)
<223> Artificially synthesized primer sequence,
5B531HRV

<223> Artificially synthesized primer sequence 5B591HRV

<400> 10
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seq list

<210> 11
 <211> 52
 <212> DNA
 <213> Artificial Sequence

<220>
 <221> primer_bind
 <222> (1)...(52)
 <223> Artificially synthesized primer sequence,
 5B591HRV

<223> Artificially synthesized primer sequence

<400> 11
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<210> 12
 <211> 8
 <212> FRT
 <213> Hepatitis C Virus

<220>
 <221> VARIANT
 <222> 1, 3
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<221> VARIANT
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 <223> Xaa = Any Amino Acid

<400> 12
 Xaa Asp Leu Ser Gly Trp Phe Xaa
 1 5

<210> 13
 <211> 8
 <212> PRT

seq list

<213> Hepatitis C Virus

<400> 13

Lys Asp Leu Ser Gly Trp Phe Lys

1

5